THE PRESENT GEOGRAPHIC DISTRIBUTION OF YELLOW FEVER AND ITS SIGNIFICANCE[1]

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IN YELLOW fever as in many other infectious diseases it is becoming evident that missed cases are the majority of the infections. Without laboratory help the numerous mild cases, as well as many atypical severe ones, could not be identified. It was not possible to devise the needed laboratory methods until susceptible animals became available, and this did not happen until a few years ago. Human volunteers made possible the carefully controlled experiments of Walter Reed at the beginning of the century, but it was not considered justifiable to continue to use man, however willing, in experiments with this dangerous disease after it had been proved that the mosquito *Aedes aegypti* was the vector and that communities had it in their power to protect themselves by controlling these insects.

There was, therefore, a long period without great advances in yellow fever research, but during this interval there was wide application of the knowledge of the mosquito vector to yellow fever control. There were conspicuous successes in cities that had frequently been visited by the disease and in tropical seaports in which the coming of susceptible foreigners had constantly fostered epidemics. With the suppression of yellow fever in cities, it seemed as a rule to disappear from the surrounding towns and

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countrysides. While this observation was doubtless correct in some regions, there are other places in which the infection must have remained prevalent in missed cases, only to reappear at times in epidemic form.

As will be brought out later, it seems that some conditions which favor the persistence of yellow fever infection in parts of South America and Africa do not exist in the more easily controlled region on the west coast of South America and around the Caribbean Sea and the Gulf of Mexico. It was within the latter general region that Henry R. Carter had most of the experiences on which he based his lucid exposition of yellow fever epidemiology; that Walter Reed showed that the aegypti mosquito was the essential vector; and that W. C. Gorgas experienced his outstanding successes in control and reached the opinion that yellow fever could rapidly be swept from the world by the same methods. It was only when the missed cases were being discovered that it was learned that the early observations do not apply *in toto* to all parts of the world.

If suitable laboratory tests for the recognition of recent and present yellow fever infection had formerly been available, the leaders in yellow fever thought and work would not have concluded that the disease depended for its continued existence on endemic foci in a limited number of key cities which could be sought out and controlled. We would not have thought in 1925 that the few recognized cases in the northeastern part of Brazil were the last manifestations of the disease before its disappearance from the Western Hemisphere. Such optimism seemed justified at the time, however, by the rapid regression of recognized yellow fever in the Americas.

THE HISTORIC DISTRIBUTION OF YELLOW FEVER

The historic records of the distribution of yellow fever were determined by the reports of cases that could be clinically diagnosed. Recognition depended primarily on the presence of several of the characteristic symptoms—sudden onset, fever, severe headache and backache, prostration, slowness of pulse in relation to temperature, black vomit and bleeding gums, pronounced albuminuria

and sometimes anuria, and a short duration. The mild and atypical cases were usually overlooked unless associated with typical severe illnesses in epidemics. In the more recent past there was a tendency to rule out yellow fever regardless of suggestive symptoms, if aegypti mosquitoes were not present. There was also unavoidable confusion with severe malaria, Weil's disease, and other affections which sometimes simulate yellow fever.

The distribution of yellow fever thus diagnosed had certain striking characteristics. It was common in foreigners in tropical seaports and river ports of South America, the West Indies, Central America, Mexico, and West Africa, and in the crews of ships that had touched in these places, and it appeared as warm-season epidemics in many temperate-zone cities visited by infected ships. The close relation of recognized yellow fever to the pathways of commerce has long been recognized, as is witnessed by the following observation of Hirsch in 1883: "Among the peculiarities of distribution which show the dependence of yellow fever upon locality, the first to arrest the attention is the association of the disease, not perhaps exclusively, yet to a very great extent, with sea-coasts and the shores of great navigable rivers."

Another characteristic of recognized yellow fever was a tendency to restrict itself to the larger communities. Hirsch says, "Another limitation to the area of yellow fever as an epidemic may be observed in the fact of its occurring almost solely in places with a crowded population; almost exclusively, therefore, in towns, and particularly in populous towns." Carter (1931) in developing this principle held that a group of small communities with close travel relations might be able to maintain "regional endemicity" when singly they would not have had enough susceptible babies and newly-arrived immigrants to keep the infection from exterminating itself through immunization of the population.

In recent times it was held by many that mild unrecognized cases in babies and very young children were the explanation of the widespread immunity of the natives and were also the hidden source of the frequently observed infection of newly arrived foreigners when there were no known previous cases. It was supposed that the disease in very young children differed from that in the

adult in being mild and without characteristic symptoms, although some observers reported fatal cases of yellow fever in children. Boyce, speaking of West Africa in 1911, emphasized the probability of wide distribution of unrecognized yellow fever in the natives and did not limit this hypothetical infection to infancy: "Then it is to the black races that we must look for the source of supply of the yellow fever virus; it is they who, in childhood and adolescence, have the disease in a mild form: but mild though it be, quite sufficient to infect the *Stegomyia*, as the inoculation experiments of the American Commission proved. In other words, the black natives have the so-called mild or ambulatory form of yellow fever. These mild forms pass unrecognized amongst the natives just as the sister disease—malaria—does, but, nevertheless, like malaria, it is there." This statement seems almost prophetic in the light of recent findings, but the close comparison with a chronic infection like malaria is unfortunate, for there is no reason to believe that a person infected with yellow fever virus ever remains infective to the mosquito for more than a few days.

A few examples (Boyce, 1911) will illustrate sufficiently the wide distribution and severity of the yellow fever epidemics in cities before the aegypti mosquito had been incriminated, in the days when wooden sailing ships were transporting yellow fever virus in man and mosquito, and breeding the vector in their water casks. Yellow fever was brought into Barcelona in 1821 by ships from Havana, and 20,000 people are said to have died of the disease. New Orleans was visited frequently by yellow fever and had an epidemic as late as 1905. The epidemic which occurred there in 1878 caused 4,046 deaths. Yellow fever has extended from New Orleans up the Mississippi as far as southern Illinois. Philadelphia also had a long list of epidemics. The most severe ones occurred in 1793 and 1803 with 4,044 and 3,900 deaths respectively. The experience of New York during a limited period has recently been summarized by Bolduan (1933). The disease was epidemic thirteen times from 1791 to 1807 and the city lost nearly a tenth of its population in consequence. The last outbreak in New York was in 1870. In the epidemic of 1798 Bolduan

estimates that 1500 deaths resulted, in a population of about 60,000. Sixteen doctors lost their lives out of an estimated total of not more than 40.

Such disasters seldom happen under present conditions. The only recent yellow fever epidemic in a large city was the one in Rio de Janeiro in 1928 and 1929, with 435 deaths (League of Nations, 1931). That such an accident could happen shows how important it is that all the silent endemic areas from which come the seeds of epidemics should be discovered and studied. This is urgent now that air travel is making it possible to go from country to country and continent to continent within the incubation period of yellow fever.

THE IMMUNITY SURVEY BY PROTECTION TEST

The first and most important preliminary step toward obtaining an immunity test for use in revealing hidden yellow fever was taken in 1927 by Stokes, Bauer, and Hudson (1928). Working in West Africa they transmitted yellow fever from man to two species of Asiatic monkeys, *Macacus sinicus* and *M. rhesus*. The rhesus monkey proved to be the more susceptible and soon came into general use in laboratory studies of yellow fever. A few years later Theiler (1930) found out that mice are susceptible to yellow fever virus, if inoculated in the brain, and observed that infection was prevented if immune serum was mixed with the virus before inoculation. The discovery of these two susceptible animals, the monkey and the mouse, opened up a great field of experimentation in yellow fever and made possible the extensive study of the geographic distribution of yellow fever immunity, now almost completed, which will be summarized in this address. This study is only one of the numerous investigations of yellow fever which have been carried out in recent years by members of the staff of the International Health Division of the Rockefeller Foundation under the central direction of Dr. Frederick F. Russell and with the coöperation of many governments. In charge of the work in West Africa was Dr. Henry Beeuwkes and the direction in South America is in the hands of Dr. F. L. Soper. Laboratory tests of specimens from other parts of the world were made here in New York by my associates and myself in the Laboratories of the International Health Division located in The Rockefeller Institute. An idea of the extent of the work can be obtained from the number of persons of many countries from whom blood has been taken for testing in the three laboratories of the Division. In round numbers up to the end of November, 1934, sera from 33,000 persons were examined for the power to protect mice against yellow fever virus—11,000 in Lagos, Nigeria, 7,000 in Bahia, Brazil, and 15,000 in New York.

The first yellow fever immunity survey was made in Nigeria by Beeuwkes, Bauer, and Mahaffy (1930). As only monkeys were then available and as it was found necessary to use two rhesus monkeys for each serum tested, the number of blood specimens which could be examined was severely limited and only a few communities could be studied. This handicap was removed, however, when mice became available.

Soon after Theiler's announcement that mice had been found to be susceptible, Sawyer and Lloyd (1931) undertook to contrive a protection test technique which would make possible the use of these animals in extensive immunity surveys. They had obtained results too irregular for this purpose when the sera were simply mixed with small amounts of virus and injected directly into the brain. A method was finally devised, the "intraperitoneal protection test in mice," which was found to be practical, sensitive, and dependable if care was taken to use only healthy mice of highly susceptible strains.

Six mice were used for each test serum and each control. Before the injection of the serum-virus mixture the mice were anesthetized with ether, and a small amount of starch solution was injected into the brain of each to localize the virus by producing a slight injury. This was necessary because yellow fever in the mouse, unlike the disease in man or monkey, is exclusively an affection of the nervous system and essentially an encephalitis, and the virus cannot pass from the circulating blood to the brain cells to produce disease unless the barrier between the blood stream and the brain is broken. Then the mixture of the test serum with a 20 per cent suspension of infective mouse brain was injected into the perito-

neal cavity. The virus used had been modified by over a hundred passages in mice and had become highly "neurotropic" with the loss of its power to attack the abdominal organs of susceptible monkeys. If five of the six mice used in the test of a serum survived for ten days and the controls with known normal and immune sera were satisfactory, the test serum was considered protective. This result was interpreted as meaning that the donor of the serum had been infected with yellow fever virus at some time in his life. If four or more of the six mice died from the fifth to the tenth days after inoculation, the result was considered "no protection," and it was held highly probable that the donor had never had yellow fever. Intermediate results were considered inconclusive, and the tests were repeated when possible. The detailed technique and method of interpreting the results are given in the original description by Sawyer and Lloyd and in the later report of Mahaffy, Lloyd, and Penna (1933) on their extensive experience with the test in Brazil and Africa.

The use of the protection test in delimiting the areas of recent yellow fever infection and in epidemiological studies was based on the observation that an attack of yellow fever is followed by an enduring immunity with protective antibodies in the blood. As a rule, these antibodies persist for life, as was shown in tests of the sera of 60 persons residing in the United States who were supposed to have had typical attacks of yellow fever many years before in the United States or the West Indies (Sawyer, 1931). Forty-five, or 75 per cent, of the sera protected rhesus monkeys against virulent yellow fever virus. One of the protective sera came from an aged woman who had had yellow fever in Louisiana 78 years before. Protection was given also by five of six sera from persons who had had their attacks 75 years before in the last epidemic in Norfolk and Portsmouth, Virginia. After making due allowances for possible mistakes in clinical diagnosis and the relative insensitivity of the protection test in monkeys in comparison with that in mice, the results suggest that demonstrable immunity in a person who has had yellow fever endures as a rule for life, but that in exceptional instances the antibody content of the serum may fall with the passage of years until the results of the protection tests in mice may be "inconclusive" or even "no protection." It is unknown whether the immunity could ever fall so far as to permit a second attack of yellow fever.

While it is of value to know that negative protection test results mean as a rule that the person supplying the serum has never had yellow fever, it is of greater importance to our studies to ascertain with what certainty we can accept protection by a serum against yellow fever virus as conclusive evidence of past yellow fever infection. Evidence of the specificity of the test was obtained in experiments with monkeys and in tests of the sera of persons after known attacks of yellow fever and also both before and after vaccination with living modified yellow fever virus and human immune serum by the method of Sawyer, Kitchen, and Lloyd (1932). While it was easy to show that the blood of persons and monkeys regularly acquires protective power after infection with yellow fever, it was more difficult to prove that the presence of protective power always means previous infection with yellow fever virus. The best evidence was obtained by examining many sera from countries or regions in which yellow fever has presumably never been present. The results have been reported by Hughes and Sawyer (1932), Mahaffy, Lloyd, and Penna (1933), and Sawyer and Whitman. In the group of countries and regions thus investigated were included Canada, China, the Philippine Islands, the Malay States, India, Ceylon, Australia, and two high-altitude areas of Brazil. In most of these places no protective sera were found. Considering the specimens as one group, we find that there was one protective serum for about every 200 specimens. False protection due to cross-immunity with any common disease other than yellow fever is rendered highly improbable by the results just presented and also by experiments of various investigators showing that sera of persons convalescent from dengue (Stefanopoulo and Callinicos, 1932; and Snijders, Postmus, and Schüffner, 1934), Weil's disease (Sawyer, Kitchen, Frobisher, and Lloyd, 1930; and Stefanopoulo, 1933a), and two African diseases associated with jaundice, "Kukuruku disease" (Beeuwkes, Walcott, and Kumm, 1931), and "diounde" (Stefanopoulo, 1933b), and a number of other diseases, do not protect against yellow fever virus. The test is obviously highly specific and suitable for our studies.

The plan for collecting sera in the immunity survey varied some-

what between countries but was most commonly as follows: Representative towns and villages were selected for investigation on the basis of interest and accessibility. Blood was taken from about 50 persons in each place, 25 adults and 25 children, selected at random without regard to previous illnesses. Sometimes only smaller numbers of suitable specimens could be secured. Great care was taken to select only donors who had never been out of the locality. Age, sex, race or tribe, and other particulars regarding the donors were recorded on a standard form. In some instances, in regions in which it was thought that yellow fever had never been present, all the specimens were taken from adults in order to obtain as much information as possible from negative results regarding the yellow fever history of the region. Where there had been known epidemics the specimens sometimes were taken only from children for the purpose of obtaining the maximum amount of information about recent yellow fever.

The blood was drawn from a vein of the arm into vacuum syringes of the "venule" type with a capacity of 30 cc. and sent to the nearest laboratory for separation of the serum and shipment in ampoules under refrigeration to the laboratory in which the tests were to be made. When the blood was collected at a great distance from a laboratory, it was sometimes found advisable to draw off the serum in the field into a second venule of smaller capacity. In certain long expeditions in the African tropics an "Icyball" refrigerator on a motor truck was utilized for storing the serum.

In certain countries, for instance in the Belgian Congo and in the eastern part of Africa, the sera were obtained for us by governmental health officials and shipped to one of the Division's laboratories for testing, and in some other countries, particularly French Equatorial Africa and French West Africa, a government representative accompanied a member of the staff of the Division and coöperated in securing suitable donors and collecting the specimens. In Brazil the survey is being made by the coöperative Yellow Fever Service maintained by the National Department of Health and the Rockefeller Foundation. Specific acknowledgment of coöperation must of necessity be left to the separate regional reports.

By classifying the donors according to age in relation to immunity, it was sometimes possible to construct a rough yellow fever history of the community and even to establish approximately the dates of outstanding epidemics. The impracticability of bleeding very young children often prevented a decision as to the presence of yellow fever during the previous five or ten years. The history of epidemics and testimony regarding observed cases sometimes helped in the interpretation of the protection test results, but they were usually not available and sometimes were unreliable.

Within the areas in which protective sera are found it is possible to obtain direct evidence as to the actual presence or absence of hidden yellow fever at the time of the investigation by the routine collection of specimens of liver tissue from the bodies of persons who have died after a febrile illness of less than ten days duration. As described by Soper, Rickard, and Crawford (1934), the method involves the organization of an extensive supervised collection service and the use of an ingenious instrument known as the viscerotome, with which appointed non-medical representatives with legal sanction can rapidly punch out small pieces of liver tissue from cadavers before the issuance of burial permits. An extensive service of this kind is functioning in Brazil and services are being organized in other South American countries. The specimens are placed in formalin solution and sent to the laboratory of the Division in Rio de Janeiro where they are examined histologically for the lesions of yellow fever. The practice is based on the conviction that the existence of yellow fever in a community will result in some fatal infections. In the examination of 28,000 liver specimens up to June, 1933, yellow fever was diagnosed in 54 cases from 43 places in which the disease was not known to be present. By the middle of 1934 the total number of liver specimens examined had risen to 46,000 and there were over 900 posts equipped to obtain liver specimens. By such means it is possible, within the area in which immunizing infections have recently oc-

curred, as shown by protection tests, to find places in which yellow fever is now occurring and where intensive investigation and perhaps control measures are specially needed.

Of the methods available for studying the distribution of yellow fever, the protection test is the most objective, but it has the limitation of being applicable only to persons over five or six years old. The histological examination of liver tissue comes next in objectivity, but necessarily involves an element of opinion in the interpretation of the lesions and is applicable only to persons who have died of an acute illness. By far the most subjective and least reliable method is clinical diagnosis, which depends on observations possible only at the time of illness. There is another method which is the most conclusive of all when positive results are obtained, but it is of limited use in distribution studies because it is applicable only to a few important individual cases. It is the recovery of the virus by transferring blood from a patient, within the first three days of suspected yellow fever, to a rhesus monkey and studying the virus in the laboratory. Each of these methods gives important information not obtainable by the others, and the most complete and conclusive investigations involve the use of them all.

AFRICA

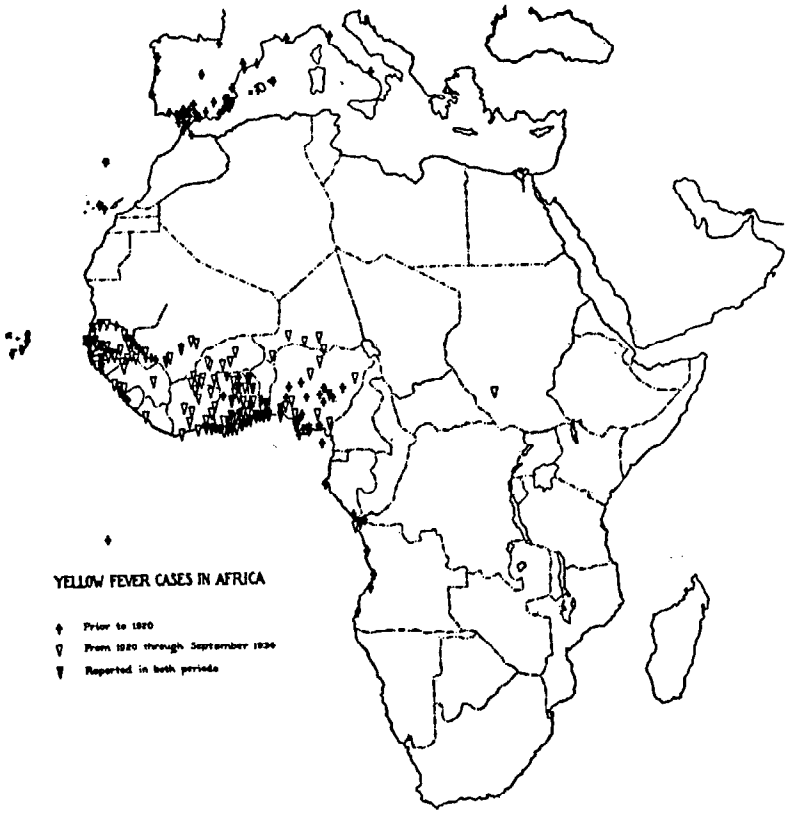
Of special interest with regard to the distribution of yellow fever is Africa. It was probably the original home of yellow fever (Carter, 1931), and it now contains one of the two great endemic regions of the world.

Carter (1931) cites records indicative of the presence of yellow fever in Africa as far back as 1585, when Drake lost two or three hundred of his men from a rapidly fatal disease soon after leaving the Cape Verde Islands for the West Indies. Since the middle of the eighteenth century there have been many reports of yellow fever from the African coast from Senegal to Angola, and in the past half-century there have been a considerable number from the navigable rivers and the railroads, particularly the railroad running eastward from Dakar. Scattered inland outbreaks have been reported also, but with a few exceptions all occurred within the

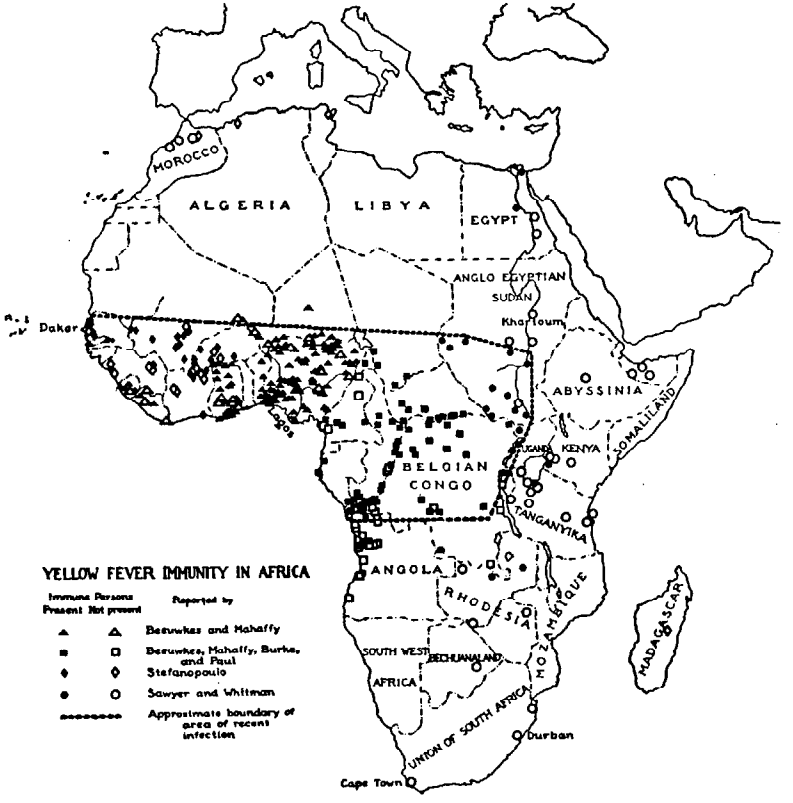
present century. Most of the cases reported were in white persons. Although the disease was believed by some to be widespread among the negro natives, it was only very infrequently diagnosed. Nevertheless, several exceptional epidemics in wholly negro towns in Gold Coast and one in Nigeria were observed and studied by Beeuwkes and his associates in 1926, 1927, and 1928. The case mortality among West African negroes is much lower than among Europeans, and it is generally considered that the negro has greater inherited resistance to yellow fever than has any other of the principal races, although he is equally susceptible to infection. This circumstance makes for subclinical or inapparent infections and accentuates the difficulty of detecting the infection among the natives of West Africa through clinical observation. As a result reported yellow fever has been confined chiefly to seaports, rivers, and railways, where there are enough susceptible white persons to reveal the presence of the virus by contracting the disease and exhibiting characteristic symptoms.

The distribution of historic yellow fever from the beginning is shown in map 1. For the collection of the published data for this map and the corresponding one for the Western Hemisphere, I am indebted to Dr. Persis Putnam. (In maps 1, 2, and 5 crowding made it necessary at times to have one symbol stand for several of the same kind.) In map 1 a distinction has been made between the cases before 1920 and the later ones. The more recent period would include the lifetime of the children being studied by the protection test and also the recent few years in which the discovery of cases has been stimulated and assisted by the immunity survey and other intensive yellow fever studies.

The systematic immunity survey of Africa began in Nigeria in 1931. The combined results of the several studies making up the survey are presented in map 2. The facts for the greater part of West Africa have been published in detail by Beeuwkes and Mahaffy (1934), and those for the French Cameroons, French Equatorial Africa, the Belgian Congo, and Angola by Beeuwkes, Mahaffy, Burke, and Paul. In the maps and tables of these publications the data for children and adults are shown separately and the percentages of positive sera are given. The remainder of



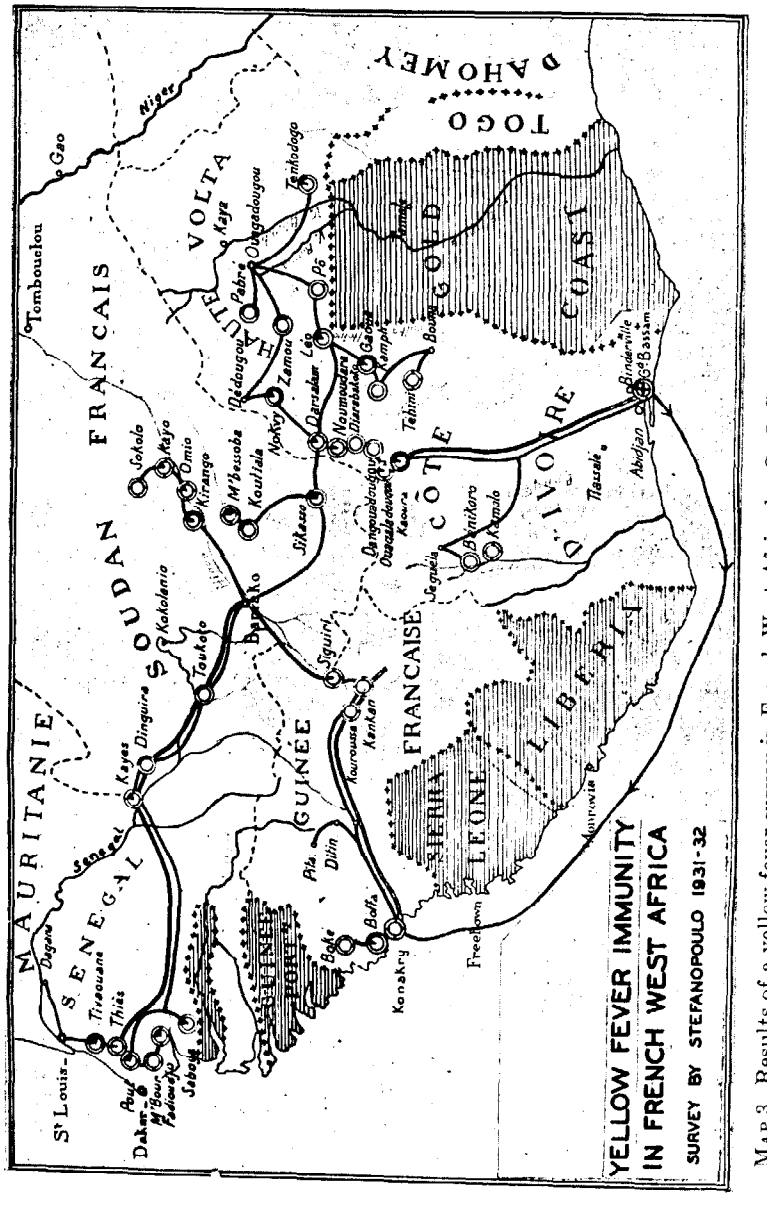
MAP 1. Places in Africa and southern Europe in which yellow fever cases were reported, classified as to occurrence before or after January 1, 1920.



MAP 2. Places in Africa classified according to the presence or absence of persons immune to yellow fever, as demonstrated by protection tests in mice in surveys from 1931 to 1934.

West Africa, consisting of a large part of French West Africa, was surveyed independently for the government by Stefanopoulo (1933a) of the Pasteur Institute in Paris and a map showing his results graphically was published by Boyé (1933). As the published map did not distinguish between the results for children and those for adults, Stefanopoulo has kindly adjusted it to agree in method with the maps of Beeuwkes and his associates and has given me permission to put it on record here (map 3). Stefanopoulo used an intracerebral protection test in mice in testing the sera which he collected in West Africa. His later tests of sera from North Africa and Madagascar (Stefanopoulo, 1934), included in map 2, were performed in uniformity with those of the other investigators, with the technique of the intraperitoneal protection test in mice. The few symbols for this last group of tests are arbitrarily placed at the capital of each country as the names of the specific locations have not yet been received. The remainder of Africa was surveyed by Sawyer and Whitman through examination in New York of sera collected and forwarded by health officials of the various governments. The completeness of the map bears witness to the excellence of this coöperation.

In map 2 the distribution of the solid black symbols, representing places in which at least one immune person was found, and the hollow symbols, showing where none of the sera gave protection, gives at a glance the general area in which yellow fever has recently been present, and within which it is probably widely but irregularly distributed at the present moment. The approximate boundaries of this area are marked by a heavy broken line. Outside the area there are a few of the solid symbols signifying the presence of at least one immune. The one in the desert north of Nigeria marks Agadez, where only one protective serum was obtained, from a child of eight years, although 50 sera from children and 36 from adults were examined. The other protective sera from outside the area, with one exception, were all from adults. The exception was the serum of a child of ten years at Golungo Alto in Angola not far from the coast. In Egypt three protective sera, one in Mansura and two in Assyut, were found among 237 from that country. In only one other place outside the area were two



MAP 3. Results of a yellow fever survey in French West Africa by G. J. Stefanopoulo. Inner circles represent children, outer circles adults. Black sectors indicate the proportion of protective sera. Circles crossed by lines signify that no tests were made. The solid line shows the itinerary (after Stefanopoulo).

positive sera obtained in one locality—Catumbela in Angola. In Kenya the one protective serum among 131 was so weak that the result was inconclusive on retest. Among 194 sera from Tanganyika, one gave protection but a second specimen taken six months later from the same donor did not. Among 96 sera from Northern Rhodesia were two protective ones from miners recruited from two different districts. South of the area in Belgian Congo there was one town in which an immune person was found, and in Angola there were two such towns in addition to the two already mentioned as sources of protective sera.

An unexpected protective serum from a community may sometimes mean that the history as to previous wanderings of the donor is incorrect, or that there is a slight content of non-specific antiviral substance in the serum, or even that there has been some error in field or laboratory. It may well be, however, that our preconceived notion of the impossibility of yellow fever in the region may occasionally be incorrect and that the donor has really undergone infection in the locality in which he is living. We must remember that the human source of the virus and the infected mosquito vectors, as well as the recipients of infection, may travel widely and might produce isolated infections in places unfavorable to the spread of the disease.

Comparison of maps 1 and 2 reveals a marked inconsistency between the distribution of reported cases of yellow fever and that of present immunity. In the region which extends from Senegal to the eastern boundary of Nigeria the discrepancy is that the numerous immunes are scattered irregularly throughout the area while the observed cases were clustered in and near the seaports or on the principal inland travel routes.

East and south of Nigeria, although yellow fever had never been reported before the immunity survey except in a few places on the Atlantic coast and on the lower Congo, the survey revealed the presence of immune persons in an area extending from Nigeria to the Nile in the Anglo-Egyptian Sudan, and from the desert in the north into Belgian Congo in the south. The number of immunes is greater in some parts of this area than in others as is shown in the reports on the separate regions. The most numerous

and most recent immunizing infections are in an area extending from Carnot, in French Equatorial Africa near the Cameroons, through Bangui and Zemio as far as Wau and Rumbeck in the Anglo-Egyptian Sudan. In strong contrast to the insignificant number of immunes found in places outside the area of recent infection (map 2) are the figures for Carnot, where the sera of 54 per cent of the adults and 32 per cent of the children gave protection and the youngest donor of protective blood was only five years old. The corresponding figures for Bangui were 32 per cent, 4 per cent, and 15 years; for Zemio, 95 per cent, 36 per cent, and 9 years; for Wau (Hewer, 1934), 26 per cent, 13 per cent, and 6 years; and for Rumbeck, 57 per cent, 4 per cent, and 11 years. As protective sera were obtained from children among the youngest tested, it is evident that yellow fever virus has been present in recent years. The percentage of protective sera is distinctly lower in the French Cameroons, the southeastern part of French Equatorial Africa, and the Belgian Congo except in the extreme north of the country and on the lower Congo. In many of the lightly immunized places no immunity in children was found. Only one inland case of yellow fever has ever been reported east of Nigeria. Following the unexpected revelations of the immunity survey a case of yellow fever in a native was diagnosed through clinical and postmortem evidence at Wau in the Anglo-Egyptian Sudan (Hewer, 1934).

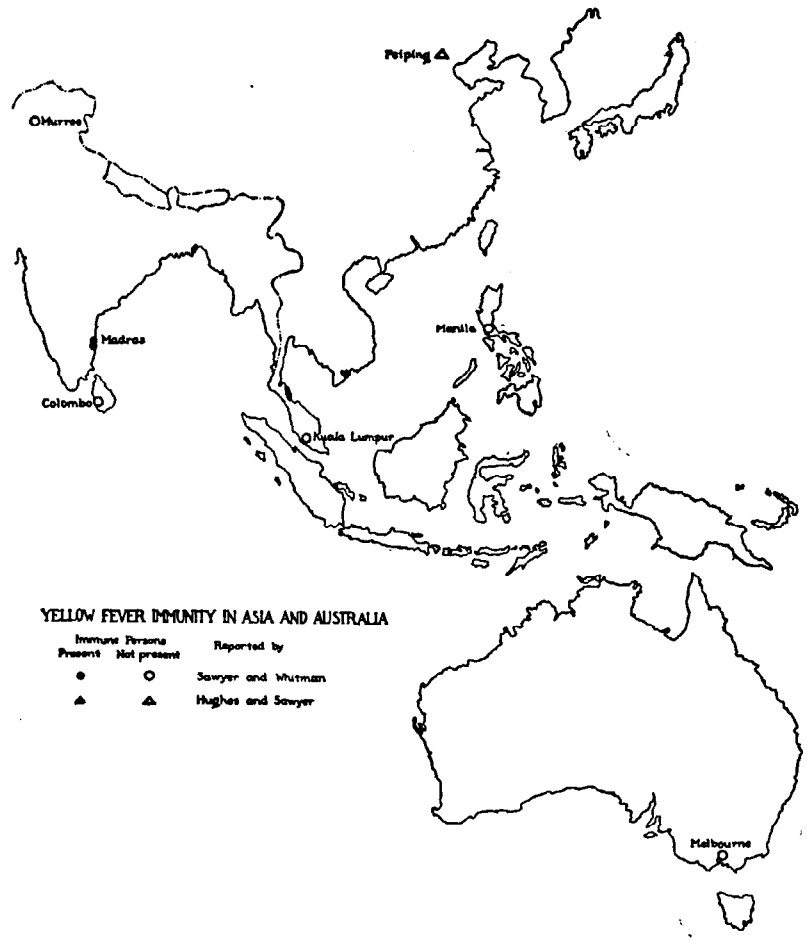
In this connection, it is of interest to read a bit of evidence presented by Carter (1931) as supporting the contention that the negro has a true racial resistance to yellow fever such as is not shown by any other race. In citing the experience of a Kordofan half-brigade sent to Mexico in the French expedition of 1862 he says: "There had been no yellow fever in Kordofan in historic times, yet these troops enjoyed an 'almost absolute immunity' and suffered no deaths from that disease, while the rest of the expedition, similarly circumstanced, suffered severely." At the time that Carter wrote, and even up to the year 1933, there was reason to think that yellow fever had never been within a thousand miles of Kordofan, but the subsequent revelations of the immunity survey showed that the infection must recently have been in that

province without establishing a recognized epidemic. The historical episode cited by Carter may now be taken as suggesting that the people of Kordofan had an acquired immunity at least as far back as 1862.

The question arises naturally whether the invisible immunizing infection may not be the work of a strain of yellow fever virus of very low virulence. It would be rash to accept this hypothesis before these silent areas have been searched much more thoroughly for yellow fever recognizable clinically and pathologically and before the virus has been obtained and studied. As will be shown later in this address, occasional fatal cases of yellow fever are discovered under somewhat similar circumstances in the Amazon Valley by examining routine pathological specimens obtained with the viscerotome.

ASIA AND AUSTRALIA

Asia and Australia have never had yellow fever so far as is known, and are practically without immune persons, as is shown in map 4. The places investigated in that part of the world were Peiping in China, Manila in the Philippine Islands, Kuala Lumpur in the Malay States, Murree, Madras, and Chingleput in India, Colombo in Ceylon, and Melbourne in Australia. Of these places the only ones in which any protective sera were obtained were Madras and the neighboring town of Chingleput. The one protective serum from Madras was so weak that the result of a retest was inconclusive, but two sera from adult residents of Chingleput gave protection in two successive tests and even when diluted. Fifteen months later second specimens were obtained for us from the same two individuals in Chingleput, and one again gave protection even when diluted, although the other then gave an inconclusive result. Sera are being collected in India on a larger scale and the donor of the one consistently protective serum from Chingleput is being investigated further. Pending the completion of this study, all that can be said is that Asia and Australia have probably been free from yellow fever during the lives of the present generation if not always, which would be in agreement with the history of those parts of the world.



MAP 4. Places in Asia and Australia classified according to the presence or absence of persons immune to yellow fever, as demonstrated by protection tests in mice in surveys from 1932 to 1934.

THE WESTERN HEMISPHERE

The Western Hemisphere has been much more widely invaded by yellow fever than the Eastern, and the experiences of its many countries, with their markedly different conditions, offer much

valuable evidence not yet satisfactorily interpreted. As shown in map 5, most of the principal seaports and navigable rivers in the tropical zone have experienced recognizable yellow fever. In many instances it was known to be continuously present for long periods. In the northern and southern temperate zones the disease was repeatedly introduced in the warm seasons and remained until interrupted by cold weather. Cities far from the tropics, for example Buenos Aires and Philadelphia, suffered devastating epidemics.

In the United States there has been no known outbreak since 1905, and no immunes have been found in that country or in Canada through the random collection of sera for protection tests in the immunity survey (map 5). A considerable number of protective sera have been obtained, however, by selecting as donors persons in the United States who had gone through typical attacks of yellow fever in the spectacular epidemics of former days.

In Mexico and Central America yellow fever epidemics have occurred so recently that it is difficult to determine with certainty that the infection is not still present. An outbreak occurred as late as 1924 in Salvador and probably originated locally from a persisting unrecognized infection. The disease was unusually active in Mexico in 1920 and was present in Tampico in 1922. With such a recent history of yellow fever, it becomes of prime importance to ascertain with laboratory help whether the disease has become extinct in these countries or is merely invisible in the quiet of a long inter-epidemic period. The protection tests have shown that there are in this general region immune children too young to have been infected in the last reported epidemics. In certain instances the evidence shows that the infection must have lingered for at least a year or two after the last recognized cases. It will be of interest in future immunity surveys to observe whether the minimum age of immune persons has risen in proportion to the number of years that have passed.

In South America the immunity survey is not yet complete, but highly significant information is available (Soper, 1934). In the shaded areas shown in map 5, immune persons in early age groups have been discovered through protection test surveys. In-

MAP 5. Places in the Western Hemisphere in which yellow fever cases were reported, classified as to occurrence before or after January 1, 1920. Places in North America, Central America, and the West Indies classified according to the presence or absence of persons immune to yellow fever, as demonstrated by protection tests in mice in surveys from 1931 to 1934. Areas in South America in which the presence of immunity to yellow fever has been demonstrated in persons of early age groups by the protection test in mice. The unshaded areas include those in which surveys have not been started, are incomplete, or have given negative results in children.

complete surveys and those which have revealed immunity only in older persons are not shown. The littoral of the Caribbean Sea and the Pacific Ocean have so far been found quite free from immunity in children, but the investigations are still under way and the situation is somewhat like that in Mexico and Central America. The coastal region from Rio de Janeiro to Parà is being protected against yellow fever through mosquito control by the yellow fever service maintained jointly by the Brazilian Government and the Rockefeller Foundation and directed by Dr. Soper. It employs over 3,000 persons, most of them uniformed inspectors. The cities and larger towns of this region are now under such complete control that aegypti larvae can usually be found in only a fraction of one per cent of the houses, with the result that the species is locally almost extinct. This work is being pushed farther and farther inland into the rural areas in which aegypti mosquitoes are present, for experience has already shown convincingly that complete control in the cities is not being followed as it was formerly believed it would be, by the spontaneous disappearance of the infection from the rural areas.

When the protection test was made available, it became possible to find out whether the Amazon Valley above Parà had actually been free from yellow fever since 1913, as the records showed. The answer is given by the shaded areas in map 5, showing that numerous children and youths born after the supposed complete disappearance of yellow fever had become immune. In the last two years it has been shown also, through the routine collection and examination of liver specimens, that yellow fever infection is occurring at the present time and is producing some fatal cases. Such routine pathological examinations are responsible for the reporting of the recent cases shown on Map 5 as occurring in the Amazon Valley, except for two frank epidemics in Bolivia and one in the center of Matto Grosso. These three epidemics were diagnosed clinically as well as serologically and pathologically and are probably secondary to the widespread silent infection. In the north, on the eastern slope of the Andes, the infected area extends into the watershed of the Orinoco. Beyond the mountain range in the valley of the Magdalena there is an

area in which there have been repeated recent epidemics, confirmed as yellow fever through the protection test (Kerr and Patiño, 1933).

From the evidence now at hand, it seems safe to conclude that yellow fever has been present in recent years in most of the coastal region of Brazil from Rio de Janeiro to Parà and is now wandering about in the Amazon Valley and in adjacent areas of the Magdalena and Orinoco watersheds, under conditions as yet unknown, and that its range includes parts of Brazil, Colombia, Bolivia, and Peru.

GENERAL OBSERVATIONS

During the survey of yellow fever immunity observations were made which help to explain the geographic distribution of the disease and throw new light on its epidemiology.

We have seen how immunizing infections may be prevalent in vast silent areas for decades without any cases being reported. In recognized epidemics as well, the missed cases may far outnumber those diagnosed, as was shown by Soper and de Andrade (1933) in an intensive study of a small outbreak in a Brazilian town. On the basis of numerous protection tests in mice, they estimated that 60 per cent of the population of over 800 was immune although there had been only 19 clinically recognized cases in the community, 13 during the epidemic and 6 before. Many undiagnosed mild illnesses occurred, however, at the time of the epidemic.

In the immunity survey it has been observed that as a rule the blood donors in the higher age groups show distinctly greater percentages of immunes than do the younger ones but that there is practically always a considerable proportion of susceptibles left. Beeuwkes and Mahaffy (1934) have published a chart showing the low start and rapid rise of the percentage of immunes with increasing ages of donors in four endemic cities of southwestern Nigeria. Muench (1934) made a statistical study of the relation of the ages of the donors to the percentage of immunes, using the combined protection test data from several small towns in the upper Amazon Valley, and obtained a cumulative picture of infection such as would result if about 3.5 per cent of the persons

remaining susceptible had been immunized each year. It may be that the higher proportion of immunes in the older age groups could in some instances be explained by a supposed rapid fall in the exposure rate in the community, such that the older persons would have been more generally immunized in their childhood than were the present children. From the abundant evidence, it seems almost certain, however, that the immunizations in the endemic or repeatedly epidemic communities are produced principally through the infection of residual susceptibles widely distributed through the different age groups. No longer does it seem reasonable to postulate that the immunizing missed infections are confined to young children incapable of showing characteristic symptoms.

It is difficult to account for the persistence and wide extent of yellow fever infection in some tropical regions in view of its easy control or spontaneous disappearance in others. Differences in meteorological conditions or density of population are not alone adequate as explanations. Among the possible influencing factors is the presence in some regions of insect vectors other than *Aedes aegypti*. It has been shown in laboratory experiments by various workers that fourteen species of mosquitoes in addition to *A. aegypti* are capable of transmitting yellow fever by bite—8 in Africa, 5 in South America, and 1 in the East Indies. The search for new vectors has become of more than academic interest since Soper, Penna, Cardoso, Serafim, Frobisher, and Pinheiro (1933) showed that a rural epidemic of proven yellow fever actually did occur in Brazil in the complete absence of *A. aegypti*. Epidemics without aegypti have since then been encountered in Bolivia and Colombia (Soper, 1934). It has also been noticed that yellow fever virus is persisting in regions with few inhabitants and little travel, and this is again stimulating curiosity as to the possibility of warm-blooded hosts in addition to man. Several species of Asiatic and South American monkeys have been shown to be susceptible to the natural yellow fever virus, and Findlay and Clarke (1934) have recently shown that the hedgehog of the Old World is also susceptible.

To sum up, there are two vast circumscribed endemic regions of yellow fever, one in Africa and one in South America. Outside of these regions epidemics are very infrequent under present conditions. The regions of endemicity include areas in which the only previously known vector, the mosquito *Aedes aegypti*, is not present, and epidemics have been observed in the absence of this mosquito. It has also been demonstrated that complete elimination of yellow fever from cities is not necessarily followed by disappearance of the infection from the tributary rural areas.

Until the factors that favor the persistence of yellow fever infection in the rural endemic regions are better known and control measures are adapted to the new conditions, there will be danger of the transfer of the virus to distant susceptible communities, especially through rapid travel. We must, therefore, for the present continue to rely for protection in part on local mosquito control in cities and towns where the aegypti could flourish and on the eternal vigilance of governmental quarantine services in preventing the introduction of infected mosquitoes and persons incubating the infection.

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